CLINICAL ORAL IMPLANTS RESEARCH

316 Posters – Material Research

Influence of anguled implant position on bone strains and stress

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Background: The angulation of immediately loaded dental implants like in the 'All-on-4' concept has a crucial effect on the initial bone stress and strain. The literature shows that standard dental implants placed at a 30° angulation cause a significant higher force on the peri-implant bone.

Aim/Hypothesis: It was the aim of this study to investigate the biomechanical behavior of different implant designs placed in different angulations and to determine the stress and strain distributions in the surrounding bone using experimental and numerical methods.

Material and methods: Two standard implants with different macro- and micro-designs and one newly developed implant design were inserted into a human mandible segment with varying angulations (straight, 15, 30°). Subsequently the experimentally analysed specimens were scanned using a μ CT and the geometry of each specimen was reconstructed three-dimensionally. The resulting models were imported into the FE package SolidWorks. The implants were loaded in-vitro with occlusal forces of up to 100 N. The non-osseointegrated state of the immediately loaded implants was simulated by performing so-called contact analyses.

Results: Measured implant deflections ranged from 5 to 35 μ m at a vertical force of 50 N. Calculated displacements were between 11 μ m and 67 μ m at a vertical force of 100 N. Maximum stresses for all implants investigated were between 3 and 72 MPa at a vertical load of 100 N. Stress spikes are located in the cortical bone in the region between implant neck and cortical bone. The design of the implant body and the macro- and microthread design clearly has an influence on the peri-implant bone stress and strain for highly angulated implants.

Conclusions and clinical implications: The biomechanical results for angulated implant placement illuminate that the implant design is a crucial factor for the peri-implant bone stress. This might be one contributing factor causing cortical crestal bone loss in the long-term for highly angulated implants (30°).

317 Posters – Material Research

Influence of different drills on the primary implant stability

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Background: The implant bed preparation has a massive impact on the primary implant stability. Depending on the bone quality the primary stability can be very different if the clinician has to use the same set of implant drills.

Aim/Hypothesis: It was the aim of this in-vitro study to develop a drill sequence for one implant system, that creates the same primary stability independent of the bone quality.

Material and methods: three different drill sequences are utilized to achieve 35 Ncm primary stability for the same implant depending on the different bone qualities D1-D4. Special foam blocks are used which simulate these different bone qualities. The clinician always starts with the drill sequence for soft bone, which is creating an under contoured implant bed preparation. Especially the apical part of the conical implant is used to create stability with the self-cutting thread design. The dimension of the following drills for higher bone qualities are closer to the final implant diameter itself. The aim is to achieve between 35–45 Ncm in every bone quality. An implant motor with an integrated torque control unit is used to determine the primary stability.

Results: The primary stability measurements ranged from 25 to 55 Ncm for all bone qualities. The mean value was 39 Ncm. Most of the implants could be placed with a primary stability between 35–45 Ncm. The design of the drill and the correct sequencing clearly influenced the primary stability.

Conclusions and clinical implications: The results indicate that it is possible to achieve a standardized predictable primary stability for one implant design in different bone qualities with a corresponding drill set.

318 Posters – Material Research

Tailored strontium-containing coatings for improved bone ingrowth deposited by means of industrial magnetron sputtering equipment

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Background: Strontium may significantly improve the healing of bone tissue by increasing the osteoblast activity, which

shortens the healing period for the patients. Moreover, strontium has been found to decrease the osteoclast-driven bone resorption. Strontium-containing coatings can ensure a localized effect where needed and can be optimally designed by means of magnetron sputtering, a physical vapour deposition technique.

Aim/Hypothesis: To identify deposition parameters and characteristics of strontium-containing implant coatings that effectively increase peri-implant bone formation *in vivo*.

Material and methods: Thin film strontium-titanium-oxide coatings grown by magnetron sputtering in an industrial-scale deposition unit were investigated. The deposition technique enables tailoring of the coating properties, e.g. chemical composition and morphology, by varying a number of different parameters such as sputtering power, deposition temperature, deposition pressure and film thickness. The coatings have been characterised by X-ray photoelectron spectroscopy, X-ray diffraction, scanning electron microscopy and Rutherford backscattering spectroscopy. Adhesion of the coatings has been evaluated by bending tests, Daimler-Benz adhesion test and electron microscopy after press fitting a coated cylinder into polyoxymethylene. Furthermore, the release of strontium from the coatings, when immersed in a phosphate-buffered saline solution, has been investigated by means of inductivecoupled plasma optical emission spectroscopy. Coatings with different strontium release profiles have been tested in two in vivo studies on implants inserted into the femur of female Wistar rats with uncoated titanium implants as reference. The minimum sample size was 10 implants for each group. Also a study on the influence of strontium-containing coatings on osseointegration in an osteoporotic animal model has been initiated.

Results: Release of strontium from the coatings can be controlled and optimized through the deposition parameters. The morphology of the coatings has proven to be a primary factor with respect to tailoring the strontium release. A good adhesion of the coatings to grade 4 titanium was achieved. The best performing coatings induced a significantly improved formation of new bone compared to the uncoated titanium references.

Conclusions and clinical implications: Strontium-containing coatings have shown to significantly improve new bone formation, and magnetron sputtering seems to be a promising technique for tailoring optimal coating properties with the use of deposition equipment directly applicable to commercial production of implants.

319 Posters – Material Research

A novel multi-phosphonated implant surface treatment for accelerated and improved bone healing: a study in sheep

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Background: Stable bone-to-implant interface and strong fixation are essential requirements for successful implant integration and patient prognosis. SurfLink[®] by NBMolecules[®] (Nano Bridging Molecules SA, Gland, Switzerland) binds covalently to titanium to form a nano-meter thin monolayer of multiphosphonated molecules on the implant surface. By virtue of SurfLink[®]'s biomimetic phosphate-like groups, the treated implant is highly hydrophilic resulting in enhanced biocompatibility. The SurfLink[®] surface treatment is designed to improve osseointegration resulting in enhanced bone to implant fixation.

Aim/Hypothesis: To evaluate how bone healing is influenced by a new multi-phosphonated surface treatment (SurfLink[®]) in an unloaded sheep model.

Material and methods: Dental implants with a roughened surface finish (SPI[®] Element, Thommen Medical) with either SurfLink[®] treatment or no treatment (control) were placed in the left and right pelvis of 24 sheep according to a well-established animal model. Animals were sacrificed after 2, 8 and 52 weeks. Overall integration of SurfLink[®] treated implants was assessed by biomechanical, histological and scanning electron microscopy (SEM) analyses at short and long-term time points. Multivariate regression analysis was performed.

Results: The torque evaluation was used as main outcome variable in the multivariate analysis. Four independent variables were found: Implant treatment (SurfLink[®] or Control), Time, Bone to Implant contact (BIC), Initial bone quality. SurfLink[®] was found to significantly increase implant fixation. The histological analysis showed further evidence of increased implant fixation. SurfLink[®] treated implants were shown to be osteoconductive already at early time points, resulting in greater bone matrix formation on the implant surface. Furthermore, SEM analysis showed that after torque testing the resulting fracture occurred within the bone on SurfLink[®] implants, rather than at the bone to implant interface as it was observed with control implants.

Conclusions and clinical implications: The results from this animal study showed that SurfLink[®] treated implants improve early and long-term implant fixation and promote faster osseo-integration. Thus in a clinical situation, enhanced early and long-term implant stability can be expected.

Loading properties of implant-abutment assemblies: an interferometric analysis – a pilot study

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Background: The increase in failure rates following the initial dental implant loading phase exemplifies the importance of undestanding implant loading properties. Implant biomechanics associates load distribution with implant size and shape. Researchers have analysed the peri-implant bone stress; however, they seldom investigated the changes in the implant-abutment assembly itself. Most of the analyses were based upon virtually simulated experiments, thus lacking empirical data pertinent for real life performance. Digital holographic interferometry is a method that enables direct insight in surface deformation upon loading in real world environment with submicron accuracy. To this date, there has not been an interferometric study that thoroughly analysed loading properties of different implant-abument assemblies.

Aim/Hypothesis: The purpose of this preliminary study was to analyse the deformation in the cervical portions of two implant-abutment assemblies under incremental axial loading by means of digital holographic interferometry and compare them based on their diameters. Different implant-abutment brands are to be analysed in the final study.

Material and methods: Experiments included a dental implant (AstraTech, Astra Tech AB, Mölndal, Sweden), represented by a 3.5 and 5.0 mm diameter implant-abutment assembly. A quasi-Fourier setup with a Spectra-Physics 25 mW heliumneon laser light source (wavelength: 632.8 nm) was used for digital holographic interferometry measurements. The object beam was expanded and collimated to reflect from a part of the base (clamping device), the exposed cervical 5 mm of an implant and the abutment. An implant-abutment assembly was preloaded with a 10 N force in the high precision implant loading device. After obtaining the first hologram, the assembly was loaded with a desired force and recorded again. Both holograms were superimposed and an interferogram was reconstructed. This procedure was repeated throughout the whole process of incremenal axial loading for the following forces: 20 N, 30 N, 40 N, 50 N, 60 N, 70 N, 80 N, 90 N, 100 N, 110 N, and 120 N. Each series of measurements was repeated three times for each assembly, with complete dismantling of the implant-loading device in between series. Statistical analysis was performed using linear mixed effect modelling in R's lme4 package.

Results: Implant-abutment assemblies exhibited a linear deformation pattern. An increase in deformation upon applying

incremental forces was significantly higher in the narrow compared to the wide implant-abutment assembly (P < 0.01). **Conclusions and clinical implications:** Digital holographic interferometry is a suitable method for load distribution analysis in implant-abutment assemblies.

321 Posters – Material Research

Zirconia, fiber and metal bar attachments: retention and strength

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Background: Retention and stability of overdentures can be obtained by metal bar attachments. Different materials used instead of metals are popular in dentistry. Especially zirconia and fiber reinforced composites are widely used in fixed prosthodontics but there is no information about zirconia and fiber reinforced composite bar attachments.

Aim/Hypothesis: The aim of the study was to compare fiber reinforced composite (FRC), zirconia and chromium cobalt (Cr-Co) bars in terms of retention force and strength.

Material and methods: An acrilic edentulous mandibular model was prepared and two implant analogue were inserted in canine area 18 zirconia, FRC and Cr-Co bars and overdenture prostheses were fabricated. All samples were stored in water for 2 weeks and then 5500 cycles of insertion and removal were applied in vertical direction. Retention force and flexural strength were evaluated using universal testing machine (Lloyd Instruments Ltd.) before and after fatigue test.

Results: Variance analyses and post hoc Bonferroni test were used to compare results (P < 0.05). 23 N, 19 N, 13 N retentive forces were found for Cr-Co, zirconia and FRC bars respectively. FRC bars showed statistically significant difference in terms of retention. 823 N, 471 N and 275 N fracture loads were determined before fatigue test for Cr-Co, zirconia, and FRC bars respectively. Resistances for Cr-Co, zirconia, FRC bars were found as 816 N, 369 N and 205 N respectively. Flexural strengths were calculated for zirconia, FRC and Cr-Co bars before and after fatigue test and found 411 MPa, 205 MPa and 960 MPa; 348 MPa, 135 MPa and 934 MPa respectively. The results showed statistically insignificant differences among all bar materials.

Conclusions and clinical implications: In conclusion, zirconia and FRC bars could be alternative materials for bar attachments instead of metal bars.

Corrosion resistance and surface characteristics of anodized biodegradable magnesium

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Background: The objective of this paper is to evaluate the surface characteristics, corrosion resistance, and cell viability of magnesium modified by plasma electrolytic oxidation (PEO) method on the additives amounts of electrolyte.

Aim/Hypothesis: The objective of this paper is to evaluate the surface characteristics, corrosion resistance, and cell viability of magnesium modified by plasma electrolytic oxidation (PEO) method on the additives amounts of electrolyte.

Material and methods: PEO was performed by DC power supply at 300 mA/cm² for 10 min with pulse current for 100 ms on time and 100 ms off time. The mixture of 1 mol/l NaOH and 0.1 mol/l Na3PO4 was prepared as an electrolyte, and 0 mol/l, 0.1 mol/l, 0.2 mol/l, 0.3 mol/l glycerol were added in the electrolyte as an additive, respectively. Surface characteristics were defined by observation of surface morphology, elements analysis, and surface roughness test. Moreover, to evaluate corrosion resistance, the potentiodynamic polarization was executed in a simulated body fluid (SBF), and the cell culture test was also performed for the cell viability.

Results: The results obtained were summarized as follow; (1) The oxide layer of MgO crystal was formed by plasma electrolytic oxidation, which was more dense and homogeneous layer with the addition of glycerol. (2) Mg, O, and P were examined by EDS analysis on the oxide layer, and the peak associated with MgO was observed by XRD analysis. (3) The crystal size and the mean roughness had different values. (4) The corrosion resistance of magnesium was enhanced by plasma electrolytic oxidation, and it was increased with addition of glycerol. (5) The cell viability of anodized group was very excellent after 3 days incubation of MC3T3-E1 cell, and the highest value was examined in group of 0.3 mol/l glycerol.

Conclusions and clinical implications: The results obtained were summarized as follow.

323 Posters – Material Research

Feasibility testing of a new abutment design

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Background: Implant treatment and its evolution is driven by for example, increased product strength, ease of use for the dentists, and shortening of treatment times, all with the aim to improve the patient's quality of life.

Aim/Hypothesis: The aim of this laboratory study was to prove the mechanical performance of a new abutment design (test A) with accompanying new instrument (test B).

Material and methods: A modification of the ISO 14801: 2007 test standard was applied in test A. The modification consisted of the use of 90° angle of the load direction (instead of 30°) of the failure testing, and mounting (with corresponding bridge screw) of a test cylinders, enabling the actual force transfer to the implant-abutment complex. The new abutment (33° UniAbutment[™] EV 4.8, DENTSPLY Implants, Mölndal, Sweden) composed the test group. This new, one-piece, abutment has a conical top of 33° allowing misaligned implants to be restored with a screw retained construction. Currently available one-piece abutments (20° UniAbutment[™] 4.5/5.0 and 45° UniAbutment[™] 4.5/ 5.0, DENTSPLY Implants, Mölndal, Sweden) composed the two control groups. Abutments were tightened to the respectively implants with 15 Ncm in the two control groups and with 25 Ncm in the test group. The corresponding bridge screws holding the test cylinders were tightened with 15 Ncm. Strength analyses were done using Wöhler curves where number of cycles and fracture loads are presented. Mechanical integrity of the new instrument (abutment driver UniDriver[™] EV) was evaluated by a pick-up, carrying, engaging and disengaging test using Driver Handle and Torque Wrench (test B). 1500 torque repetitions of 25 Ncm and 10 torques repetitions of 100 Ncm were performed on 20 samples each. Descriptive statistics was calculated and presented. Results: Result from test A showed that the test abutment was at least 30% stronger than the control abutments, at 100,000 cyclic loads. This corresponds to approximately 50% better resistance to bending moments built into the test abutment compared to the control abutments. Results from test B showed that all drivers passed the mechanical handling test conditions. Conclusions and clinical implications: It was concluded that the new, 33° UniAbutment EV, shows superior mechanical integrity compared to both control groups (20 and 45° UniAbutments). The new instrument UniDriver EV, resisted the recommended torque levels set in the study. Hence, a one abutment solution is presented and the strength at use is granted.

324 Posters – Material Research

Digital radiographic bone density evaluation of two different implants placed in standardized porcine bone cylinders of high and low density

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Background: Implant insertion creates a lateral compression in the bone. The success of this adaptation, however, depends on several factors, including the quality and quantity of the host bone, the implant geometry and the surgical site preparation technique: the interplay of these parameters determine the initial stability of the implant.

Aim/Hypothesis: This study comparatively evaluated digital radiographic bone density adjacent to two different implants, placed in standardized porcine bone cylinders of high and low density.

Material and methods: Porcine bone cylinders were removed from the femur head (high density bone - HDB) or from the mandibular condyle (low density bone - LDB). Digital radiographs were taken from each cylinder, using a digital sensor (RVG Trophy®). Then, twenty Neodent® implants of two different models were divided in four groups: Group 1 (G1) - 5 DriveCM implants placed in 5 HDB cylinders; Group 2 (G2) - 5 DriveCM implants placed in five LDB cylinders; Group 3 (G3) -5 AlvimCM implants placed in five HDB cylinders; Group 4 (G4) - 5 AlvimCM implants placed in five LDB cylinders. All implants were inserted at bone level. After that, digital radiographs were taken from each cylinder with the implant, as described before. Two standardized regions of interest (ROIs) were defined and for each cylinder, with 1 mm in width and 3 mm in height, starting 3 mm below the top of the bone cylinder. The ROIs were immediately adjacent to the end of the threads of implants (in the left and right size of the image). The radiographic density of each ROI was calculated, and the average of the two ROIs was assumed as the radiographic density of the bone cylinder (XRD) with implant. The same calculations were made in the cylinders without implants, by superposing the digital radiographic image to the pre-defined ROIs. The preand post-implant values of each group were compared.

Results: The mean \pm standard deviation of XRD values for pre- and post-implant insertion were: G1pre = 106.2 \pm 5.9, G1post = 181.4 \pm 29.8; G2pre = 94.4 \pm 8.8, G2post = 193.4 \pm 68.4; G3pre = 105.6 \pm 4.0, G3post = 167.6 \pm 28.6; G4pre = 94.6 \pm 8.3, G4post = 171.2 \pm 50.9. There were statistically significant differences between pre- and post-values for all groups (ANOVA + Tukey tests, *P* < 0.05), but not for comparisons between implant types (DriveCM vs. AlvimCM) in the same type of bone (Friedman test, *P* > 0.05).

Conclusions and clinical implications: All groups showed a high bone compression during insertion, evidenced by increased radiographic density bone values post-implant insertion, achieving high XRD values even in LDB, without statistical differences between the two implant models (DriveCM vs. AlvimCM).

325 Posters – Material Research

Safety and efficacy of multi-phosphonate treated dental implants (randomised trial): 1 year postloading report

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Background: Improving biocompatibility of an implant surface is a long standing challenge. Several approaches have been developed over the years ranging from mechanical (e.g. blasting, machining) to chemical (e.g. anodisation and acid etching) treatments. SurfLink[®] surface treatment by NBMolecules[®] is made up of a monolayer of permanently bound multi-phosphonated groups mimicking the surface of hydroxyapatite. The SurfLink[®] surface treatment is designed to improve osseointegration resulting in enhanced bone to implant fixation.

Aim/Hypothesis: To evaluate safety and clinical efficacy of the SurfLink[®] surface treatment.

Material and methods: Twenty three partially edentulous subjects requiring at least 2 single implant-supported crowns were randomised according to a split-mouth design to receive one test (SurfLink[®] treated) implant and an identical non-treated control implant. Implants used were cylindrical titanium grade IV sand-blasted and acid etched implants with internal connection (SPI[®] Element, Thommen Medical). If more implants were needed, test implants were placed. Mandibular implants were submerged for 3 months and maxillary for 6 months and then loaded with definitive single crowns. Outcome measures were implant success, marginal peri-implant bone level changes, complications and adverse events, and marginal bleeding. The study was designed as quadruple blinded. Paired *t*-tests were used to calculate the difference of continuous outcomes between the two groups. The trial was carefully monitored.

Results: Up to 1 year after loading, one patient dropped-out, minor temporal deviations from the protocol were observed but no implant failures, complications and adverse events related to the implants occurred. No bleeding was observed when running a periodontal probe in the peri-implant soft tissues. The study is still blinded. At implant placement and all following time periods to date, marginal bone levels were comparable between groups (Table 1). Only small differences were observed between the two groups at implant loading (P = 0.76), three months after loading (P = 0.66) and 1 year

	Implant placement (Baseline)	Loading	3 months post-loading	1 year post-loading
	Mean (SD)			
Type 1 implants Type 2 implants	-0.73 (0.68) -0.88 (0.61)	-1.81 (0.73) -1.88 (0.79)	-1.83 (0.70) -1.90 (0.69)	-2.15 (0.73) -2.03 (0.78)

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after loading (P = 0.057). Table 1: Mean radiographic periimplant marginal bone levels between implant types and time periods. N = 22.

Conclusions and clinical implications: These preliminary results show that the implants with a monolayer of permanently bound multi-phosphonated groups (SurfLink[®] surface treatment) are at least as effective and safe as conventional control implants.

326 Posters – Material Research

Cell adhesion on titanium surface coated with human type I collagen

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Background: Coating titanium surfaces with extracellular bone matrix components has been often investigated to enhance initial bone healing. Type I collagen is the main organic component of bone matrix and provides a structural framework for cell adhesion and new bone formation.

Aim/Hypothesis: The aim of this study was to evaluate cell adhesion and spreading on moderately rough titanium surface coated with human type I collagen.

Material and methods: Human type I collagen was covalently linked to the surface of moderately rough (Sa $1.0 \pm 0.1 \ \mu m$) grade 4 titanium discs $(12.7 \times 2 \text{ mm})$ using polyethylene glycol (PEG) as spacer molecule and carbodiimide as catalyst. For this, the discs were immersed in a solution of diacid-PEG (0.12 mol/l) and carbodiimide (0.12 mol/l) in D-PBS for 3 h at room temperature and followed by immersion overnight in solution of human type I collagen (0.003%) at room temperature. The collagen-coated discs (CC) were used immediately and compared to the control group (TT), which did not received any coating. The collagen scaffold was evaluated by atomic force microscopy and x-ray photoelectron spectroscopy. To evaluate the cell response to the collagen coating, SAOS-2 osteoblasts $(2 \times 104 \text{ cells})$ were seeded on each disc (n = 6)and cultured for 24 h. The initial cell adhesion and proliferation were evaluated by tetrazolium colorimetric assay. Cell morphology was investigated by scanning electron microscopy (SEM) and confocal laser scanning microscopy (DRAC5 and phalloidin dyes).

Results: CC surface exhibited small collagen filaments on the surface associated with higher organic elements (N, C and O) than the TT surface. CC group exhibited more cells attached after 24 h when compared to TT group (P < 0.05). SEM and confocal observation revealed more lamellipodia and filopodia among the cells adhered on CC group.

Conclusions and clinical implications: Collagen coating can improve the activity of human osteoblasts.

327 Posters – Material Research

The risk of Schneiderian membrane perforation during sinus floor elevation procedure due to the diameter of biomateriials

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Background: Sinus floor elevation procedure gives possibility of full implant rehabilitation in the lateral region, where the bone level is not adequate. The main risk of failure this procedure is the perforation of the Schneiderian membrane.

Aim/Hypothesis: The aim of the study was to assess the risk of Schneiderian membrane perforation during application of biomaterials to the maxillary sinus due to the diameter of the granules.

Material and methods: Schneiderian membrane elevation procedures were carried out on specially prepared animal specimens (pig's head). Before every surgery, thickness of osseous tissue in the place where implant is to be located was measured by means of an adjusted and calibrated caliper. The sinus flor elevation was done by hydrokinetic method with Cas - Kit. After lifting the Schneiderian membrane 0.5 g of one of two studied biomaterials, different from one another by the diameter of the granules, was applied to the maxillary sinus. 10 surgeries were carried out using Bio - Oss of 0.25-1.0 mm diameter and 10 using Bio - Oss of 1.0-2.0 mm diameter. After that the implants of 4.5 mm diameter and 13 mm length were placed. After every procedure a sample of mucus membrane lying directly over the applied material was collected. Obtained material was studied under scanning electron microscope.

Results: No Schneiderian membrane was perforated during sinus floor elevation procedures when Bio – Oss of 0.25–1.0 mm was used. When Bio – Oss of 1.0–2.0 mm diameter was used mucosa membrane perforation occurred in two cases, both during material condensation

Conclusions and clinical implications: When studying mucus membranes, which were in contact with Bio – Oss of 0.25–1.0 mm diameter, under scanning electron microscope it could be observed that the whole surface looks very similar, it means that the material localization inside maxillary sinus was even and force applied during condensation spread steadily. When mucus membrane which was in contact with Bio – Oss of 1.0–2.0 mm diameter was studied it could be observed that they were damaged during material condensation. Because of bigger diameter granules localization inside maxillary sinus is uneven, therefore force applied during condensation influence smaller surface, which poses the risk of perforation and micro cracking of Schneiderian membrane.

Effect of copper ions on the growth of BMSC on porous HA/TCP scaffolds for vascularization of bone tissue engineering

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Background: Different approaches have been investigated to develop vascularisation in biomaterials. Copper is known to stimulate endothelial cell proliferation and enhance angiogenesis *in vitro* and is particularly involved in the activity of several transcription factors (via HIF-1 and proline hydroxylase) and bind to cell membrane releasing complex, facilitating release of GFs and cytokines from producing cells.

Aim/Hypothesis: The purpose of this study was to introduce copper into HA/TCP, one kind of biodegradable inorganic ceramics, and then copper would be released during the degradation of materials. According to the cellular biocompatibility study, it resulted to the optimal copper dose in copper-doped HA/TCP, and its potential to support the growth of BMSCs for bone tissue engineering.

Material and methods: copper-doped HA/TCP scaffolds with different doses of copper (0 μ mol/l, 3.125 μ mol/l, 6.25 μ mol/l, 12.5 μ mol/l, 25 μ mol/l) and 10 samples of each group were prepared, and then analyzed by scanning electron microscopy (SEM), X-ray diffraction (XRD) and Thermmogravimetry (TG). The effects of copper-doped HA/TCP on cells' proliferation and differentiation were evaluated by MTT and ALP activity assay.

Results: The results showed that porous HA/TCP did not exert cytotoxic effect on the cells. the proliferation and differentiation of the growth of BMSCs on the HA/TCP containing a dose of 6.25 µmol/l copper showed a higher level compared to other groups, which was optimal according to the results of MTT and ALP activity assay. In addition, the cells on the copper-doped porous HA/TCP (6.25 µmol/l) formed a continuous layer on the outer and inner surface observed by scanning electron microscopy (SEM) and confocal laser scanning microscopy (CLSM). The results suggested that the biodegradable HA/TCP could stimulate the proliferation and differentiation of BMSCs in vitro after addition of proper dose of copper. Conclusions and clinical implications: copper-doped HA/TCP scaffolds may have synergistic effects which are highly attractive in the fields of tissue engineering (e.g., bone) and biomaterials. This may allow clinical repair of large bone defects with tissue engineering techniques.In addition, it may help realize better osseointegration of implant.

329 Posters – Material Research

Thermal effects during implant site drilling comparing the movement and irrigation system

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Background: One of the issues that can contribute to a successful osseointegration of dental implants is to perform a surgery that should be less traumatic as possible. While preparing implant site, the overheating of surrounding bone due to attrition of drills while rotating can cause a local bone necrosis, influencing biological stability through the deterioration of the organic portion of the bone tissues. This situation can have a direct implication in osseointegration process, influencing peri-implant bone loss rate and implant survival.

Aim/Hypothesis: The purpose of this study was to compare the temperature variation during implant osteotomies with the external irrigation technique and double irrigation with continuous and intermittent movement.

Material and methods: Two drills (Implacil De Bortoli, Sáo Paulo, Brazil) were tested. Ten bovine ribs were used for drilling procedures with an experimental computed machine, measuring the maximum temperature in the cortical bone during the osteotomies to the preparation of a surgical bed for the installation of dental implants, with a depth of 10 mm. Two control groups without irrigation: No irrigation and continuous movement (Con1); No irrigation and intermittent movement (Con2). Four experimental groups: External irrigation and continuous movement (Exp1); External irrigation and intermittent movement (Exp2); Double irrigation and continuous movement (Exp3); Double irrigation and intermittent movement (Exp4). Twenty drillings were performed for each group.

Results: The results presented statistically significant differences between the groups (P < 0.05), with the greater efficacy in the technique of double irrigation and better performance with intermittent movements (average = 62% less). The average of thermal increase in the groups was: 8.81 ± 1.20°C in Con1 group; 6.55 ± 0.92°C in Con2 group; 4.77 ± 1.53°C in Exp1 group; 2.59 ± 0.81°C in Exp2 group; 1.37 ± 0.58°C in Exp3 group; 0.82 ± 0.36°C in Exp4 group.

Conclusions and clinical implications: The technique of double irrigation resulted in a smaller increase of temperature in the cortical bone, in both drilling movements performed, demonstrating its larger efficiency and may be more beneficial when compared to the technique of external irrigation.

Aspects of the microbial biofilms formation and development on inert substrates in implantprosthetic rehabilitation

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Background: The electron microscopy applied to the surface of medical devices or tissue taken from cases of chronic infections not associated with medical devices demonstrated the presence of bacterial biofilms, including bacteria embedded in a matrix of exopolysaccharides. Microbial biofilms develop preferentially on inert surfaces or death tissue and often appear on medical devices.

Aim/Hypothesis: The aim of this paper is to study the formation and adhesion ability of microbial biofilms on the surface of materials (inert substrates) commonly used in implant-prosthetic rehabilitation.

Material and methods: In order to achieve this study were used 40 microbial aerobic strains and 40 anaerobic microbial strains, isolated from the supragingival plaque, taken from a total of 28 patients implant-prosthetic rehabilitated, during 2011–2013. The tested materials were impression materials most commonly used in implant-prosthetic rehabilitation: irreversible hydrocolloids (with or without antiseptic substances incorporated into their structure), addition silicone, condensation silicone and polyether. The test for the ability to form biofilms on different substrates used in the clinical practice, was conducted by inoculating the suspensions obtained from cultures in logarithmic stage of development in simple broth plus a sample of test material. Samples were incubated for 24 h at 37°C.

Results: Microscopic examination of strains isolated on different solid culture media by Gram staining revealed a diverse microbiota with different morphologies: cocci Gram-positive, Gram-positive cocobacili, Gram-negative and Gram-positive yeasts and actinobacteria. Tests for the ability to form biofilms on different substrates used in the clinic showed that among the materials tested, addition silicone samples were the least colonized microbial samples followed by condensation silicone. The most colonized materials were the irreversible hydrocolloids and the polyether.

Conclusions and clinical implications: These results suggest that microbial biofilms formed on the surface of the foregoing materials, may be involved both in the etiology of oral cavity infections, and in the production of subsequent infections for both patients and the medical team (prosthetic specialist, dental nurse, dental technician).

331 Posters – Material Research

How to rationally improve a dental implants system: an example from Brazil

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Background: The majority of dental implant systems developed off-axis usa/europe, are presented as copies of legacy systems. For many years, such an attitude, has kept these markets and patients, one step behind the latest technology. However, with significant investments in research and development, some of these companies succeeded in developing systems with their own characteristics.

Aim/Hypothesis: The objective of this paper is to present how a brazilian dental implants companie has revolutionized their system along 3 years, based on scientific research. All studies that supported the changes and updates in these dental implants were properly accepted for publications in respectable journals by the international scientific community. The presented data corroborate with the trends and principles of contemporary implant dentistry. And also, can be used as starting points for future studies.

Material and methods: It will be presented the results of five different studies *in vitro* and *in vivo*. The topics include surface treatments, surgical instrumentation, prosthetic connections, the implant design and the material of implant composition.

Results: The summary of results is shown in A table, and will be properly detailed orally.

Conclusions and clinical implications: This sequence of investigative studies demonstrated the need for continued investment in research and development on the part of biomedical industry. Also, as a final result, brought to consumers an innovative product with unique features and better biomechanical behavior. This brand new system, according to a new philosophy, was properly logged along the regulatory agencies of that country before being marketed.

332 Posters – Material Research

Optimization of preload and torsion by using a unique abutment screw design for each implant platform size

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Background: When an abutment screw is tightened a preload is created securing the seating of the abutment in the implant. The tightening also induces torsion stresses to the screw shaft, increasing the risk for screw loosening. Hence, it is important to achieve sufficient preload while at the same time minimize the amount of torsion in the screw. **Aim/Hypothesis:** To justify an implant system where a unique abutment screw design applies to each implant size and where the same insertion torque applies for all abutments.

Material and methods: Two implant sizes (OsseoSpeed[™] 3.5/ 4.0 and 4.5/5.0) with corresponding abutment screws (M1.6, M2.0) were used as controls. To these controls, TiDesign[™], ZirDesign[™] and ATLANTIS[™] abutments were connected (all from DENTSPLY Implants, Mölndal, Sweden). The test group consisted of the further developed implant system Osseo-Speed[™] EV 3.0, 3.6, 4.2, 4.8 and 5.4 (DENTSPLY Implants, Mölndal, Sweden) with corresponding abutments and unique color coded abutment screws; M1.4 (Green), M1.6 (Purple), M1.8 (Yellow), M2.0 (Blue), M2.0 (Brown), respectively. All abutment screws were torqued to 55 Ncm with a speed of 2 RPM. The torque and preload in both groups were constantly recorded with 100 Hz using Instron 55 MT torsion testing device. For the specific preload, the ratio between shear and tensile stress and the utilization factor was calculated.

Results: The preload at the recommended installation torque for OsseoSpeed 3.5/4.0 (20 Ncm), OsseoSpeed 4.5/5.0 and OsseoSpeedTM EV 3.0, 3.6, 4.2, 4.8, 5.4 (25 Ncm) was identified between 245–390 N. The ratio between shear and tensile stress was calculated and the abutment screws for the Osseo-Speed EV pillars showed an increased preload and a reduced ratio at recommended installation torque compared to the screws in the control pillars. The utilization factor ranged between 37–69% depending on the abutment screw size.

Conclusions and clinical implications: At recommended torque, all OsseoSpeed EV abutment screws received a preload above 250 N, which was above the preload in the control group. The results indicate that the magnitude of shear stress of the OsseoSpeed EV abutment screws was reduced, and the torque generated increased the preload compared to the control group. Hence, the risk for screw loosening is reduced. The utilization factor indicates how much of the screw yield strength that is 'used' and a factor close to 100% may increase the risk of implant-abutment pillar complication. However, this study showed that OsseoSpeed EV abutment screws have a safety margin of 2.2 against fractures.

333 Posters – Material Research

Bone regeneration of rat critical size calvarial defect with large-size fully inter-connected porous apatite/collagen composite

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Background: Autogenous bone graft has been yet regarded as the gold standard for bone augmentation technique and new osteo-conductive bone substitute materials should be developed.

Aim/Hypothesis: We spotlighted a noble commercial particulate apatite that had large dimension of 0.5 to 1.0 mm length

and fully inter-connected porous hydroxyapatite ceramics (IP-CHA). In clinic, particle drop-out, however, hinders the cure. To offset such drawback and provide formability, IP-CHA was mixed with porcine type I collagen. The purpose of this investigation was to examine re-generation of rat cranial critical-size bone defect by new porous apatite/collagen composite.

Material and methods: We self-prepared collagen control material (C) and porous apatite/collagen composite material (AC) by freeze-drying and de-hydrothermal cross-linking techniques and punched out to the size of 6×1 mm; and implanted them in cranial critical-size bone defects (6 mm) of 10 weeks Wistar rats for 1 day, 4 weeks and 8 weeks (n = 3 for each material and feeding period). After sacrifice, we examined bone forming trends of C and AC with micro-focus X-ray computed tomography (Micro-CT); and evaluated new bone formation by AC in defects of rats visualized with Villanueva stain liquid and doubled-labeled with tetracycline and calcein using laser fluorescent microscopy. Statistical analysis was conducted by Student t test.

Results: Micro-CT tests and ImageJ analyses clarified that the defect zone implanted with C had the mean opacity value of 72 at 1 day, while defect zones had the higher opacity values of 100 and 97 (P < 0.05) at 4 and 8 weeks, respectively. The defect zone implanted with AC had the mean opacity level of 124 at 1 day, while the opacity levels significantly increased with the addition of feeding periods from 4 to 8 weeks to 153 for 4 weeks and 174 for 8 weeks, respectively (P < 0.05). Laser fluorescent microscopy revealed that AC considerably fragmented/absorbed in the defect site, and exerted excellent osteo-conductive effect in the tissues adjacent to remaining apatite particles. Double staining vividly displayed dynamic new bone formation.

Conclusions and clinical implications: Newly prepared porous apatite/collagen composite (AC) appeared to be useful new bone substitute material, applicable to dental implantology. It is emphasized here that this composite provided fairly quick (8 weeks) cranial bone re-generation property.

334 Posters – Material Research

Shear-bond strength to resin cement after the application of nano-structured alumina coating on the surface of Y-TZP

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Background: The purpose of this study is to evaluate whether the application of nano-structured alumina coating on the surface of yttria partially stabilized tetragonal zirconia ceramics (Y-TZP) will improve resin-bond strength by increasing the micromechanical retentions on the surface.

Aim/Hypothesis: The purpose of this study is to evaluate whether the application of nano-structured alumina coating on the surface of yttria partially stabilized tetragonal zirconia

ceramics (Y-TZP) will improve resin-bond strength by increasing the micromechanical retentions on the surface.

Material and methods: A total of 80 disc-shaped specimens (15.0 mm in diameter and 2.75 mm thick) were produced, and divided into four groups depending on the following treatments: A group - air abrasion, R group - air abrasion + RocatecTM (3M ESPE), PC group - polishing + nano-structured alumina coating, AC group - air abrasion + nano-structured alumina coating. An alumina coating was created by using the hydrolysis of aluminium nitride (AIN) power and a thermal treatment at 900°C. Coating characterization was observed with Field-emission Scanning Electron Microscopy (FE-SEM). Composite resin blocks were fabricated with a composite resin Filtek Z250(3M ESPE, St.Paul, USA). After the composite blocks were bonded to each zirconium oxide ceramic surfaces with Rely X Unicem (3M ESPE, USA), the shear-bond strength was tested. The specimens in the four groups were tested for the shear-bond strength before and after thermocycling (TC).

Results: The FE-SEM analyses revealed that the application of an alumina coating to YTZP ceramics created a highly retentive surface for resin penetration. The coating showed good surface coverage. The shear bond strength to the groups PC, AC was significantly higher than to the groups A and R and both before and after thermocycling (TC). The specimens in the A and R groups showed significantly lower shear-bond strength, while the specimens in the PC and AC groups did not show any significant reduction in shear-bond strength.

Conclusions and clinical implications: The results of this study indicated that the application of nano-structured alumina coating on the surface of Y-TZP ceramics significantly improved the shear-bond strength to resin cement. The method is relatively simple and has the potential to become an effective conditioning method for zirconia ceramics.

335 Posters – Material Research

surface modification of pure titanium by a double treatment of nanotube formation and dopamine coating

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Background: Surface modification is a promising method to impart biological functionality on metals for biomedical applications including dentistry. Surface modification methods provide an opportunity to change the surface composition, structure and morphology of a material without affecting its mechanical properties. Moreover, the modified surface enables immobilization of proteins to regulate higher levels of cell function such as, the control of growth or differentiation.

Aim/Hypothesis: The purpose of this study was to investigate the effect of dopamine for nanotubular titanium to improve their bioactivity and biocompatibility.

Material and methods: Dopamine was coated to the surface of the nanotubular titanium surfaces by dipping into 2 mg/ml dopamine solution in 10 m mol/l Tris-HCl buffer at pH 8.5. All specimens were investigated surface roughness, wettability, cell growth and toxicity, and corrosion resistance.

Results: The dopamine-coated titanium was successfully prepared from dopamine solution and, bright dopamine granules were randomly distributed on titanium surface after coating process. Apatite formed around the polydopamine particles after 5-day immersion in SBF. The surface roughness values of the dopamine-coated specimens were higher than those of the pure specimens. However there is was significant difference among the groups (P > 0.05). The wettability of the dopaminecoated on anodized specimen was 22.61°, which lower than the contact angles of the pure and nanotube-treated specimens. The morphology of cells grown on the dopamine-coated specimens and negative control group were similar cell survival. As MTT test result, all treated specimens were higher than nontreated specimen for biocompatibility. The corrosion resistance of the dopamine-coated specimen was significantly higher than the non-treated specimens in both of SBF and 0.9% NaCl

Conclusions and clinical implications: Findings from this research suggested that dopamine coating offer a versatile approach for the titanium surface modification. Thus, this approach will be useful in dental applications that require precise control of cell-material interactions.

336 Posters – Material Research

The Effect of GRGDS peptide coating onto titanium dioxide nanotube of chemical immobilized method

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Background: The Arg-Gly-Asp (RGD), which is one of the biocompatible materials, is a main sequence of amino acids of integrin which take part in attachment of cell and extra cellular matrix proteins. The RGD peptide promotes cell adhesion when fixed to surface. Grafting of Gly-Arg-Gly-Asp-Ser (GRGDS) peptide of RGD molecule is biomaterial and reportedly effective on osteoblast adhesion, proliferation, and differentiation.

Aim/Hypothesis: The aim of this study was to evaluate the effect of GRGDS peptide coating onto Titanium Dioxide (TiO2) nanotubes by using chemical immobilized method, which in turn can change surface characteristics and affect the adhesion, proliferation, and differentiation of human osteosarcoma (MG-63) cells.

Material and methods: The Commercially pure titanium (ASTM Grade II, Kobe Steel, Japan) discs, 10 mm in diameter

and 3 mm in thickness, were used for the anodization process. The specimens were divided into two groups; TiO2 nanotubes and GRGDS-TiO2 nanotubes. Cell culture is carried out using human osteosarcoma (MG-63) cell. After 2 h and 24 h of cell culture, the morphology was observed by field emission scanning electron microscope (FE-SEM). Cell proliferation was examined using the EZ-Cytox assay kit at 2 and 4 days. Cell differentiation was measured using the ALP activity assay at 4 and 7 days. The surface characteristics of GRGDS peptide coated on TiO2 nanotubes were observed using the X-ray photoelectron spectroscopy (XPS). The data were statistically analyzed with independent t-test.

Results: These results were confirmed that GRGDS peptides were well coated on the TiO nanotubes surface, because of nitrogen up-regulation or C=O carbons presence. Cell proliferation of the GRGDS peptide coating group were significantly higher than TiO2 nanotubes group. ALP activity showed no significant difference between TiO2 nanotubes and GRGDS-TiO2 nanotubes group.

Conclusions and clinical implications: These results suggest that more successful osseointegration of implant can be enhanced by coating GRGDS peptide to TiO2 nanotubes.

337 Posters – Material Research

The evaluation of antibacterial activity and osteoblast viability of Titanium nitride/ Zirconium nitride coated on titanium

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Background: In terms of long-term success of the implant, periimplantitis has become a big challenge. The research to prevent peri-implantitis is rare with surface treatment. Physical modification such as coating with Titanium nitride (TiN) or Zirconium nitride (ZrN) on the implant surface was reported to reduce bacterial adhesion and to promote the growth of fibroblasts (Birte Groessner-Schreiber et al. 2001–2006).

Aim/Hypothesis: The aim of the present study is to evaluate bacterial colonization and osteoblast viability with various TiN /ZrN coating on titanium.

Material and methods: Commercially pure titanium discs (ASTM Grade II, Kobe Steel, Japan, 15 mm in diameter and 3 mm in thickness) were used as the substrate for coating with Arc ion plating system (ATS-MC-STD-300[®], A-tech system, Korea). Discs were divided into six groups (MA-Ti, TiN;99.99 wt%Ti, ZrN;99.99 wt%Zr, (Ti20Zr75Ag5) N, (Ti35-Zr60Ag5) N, (Ti47.5Zr47.5Ag5) N), according to ratio of TiN and ZrN coating on titanium. The Surface analysis was performed with contact stylus profilometer, field emission scanning electron microscope (FE-SEM) and x-ray photoelectron

spectroscopy (XPS). Colony forming unit (CFU) was measured with Porphyromonas gingivals. XTT assay was measured to evaluate of biocompatibility with human osteoblast cell line MG63.

Results: Colonies of porphyromonas gingivals on the surface of nitride coating were significantly decreased compared to control (P < 0.05). Viability of MG63 cells on the (Ti20Z-r75Ag5) N specimen was significantly higher than the control group (P < 0.05).

Conclusions and clinical implications: TiN/ZrN coating on titanium discs showed antibacterial property and biocompatibility with osteoblast. Proper ratio of TiN/ZrN coating on titanium could be a novel surface treatment prevent peri-implantitis and promote better osseointegration.

338 Posters – Material Research

Development of novel osteogenic bone substitute material using statin-loaded biodegradable polymer-calcium phosphate nanoparticle composite

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Background: Calcium phosphate (CaP) biomaterials such as hydroxyapatite (HA), beta-tricalcium phosphate (β -TCP), or their composite are commonly used as bone substitute materials. Resent researches indicated that CaP materials with porous structure exhibited the excellent biocompatibility, however, the drawback of porous CaP materials is that they are extremely brittle and fragile. We developed the novel materials with interconnected pores which had CaP nanoparticle-dispersed poly-L-lactic acid (PLLA) beams. Hydroxymethylglutaryl coenzyme A reductase inhibitors, so-called statins, are a group of drugs for the treatment of hyperlipidemia, and their osteogenic effects have also been reported. In the present study, we examined the fluvastatin-loaded PLLA-CaP composite as a bone substitute material on the promotion of osteogenesis inside and around the composite.

Aim/Hypothesis: The aim of this study is to investigate the effectiveness of the PLLA-CaP composite with statin as the osteogenic bone substitute material.

Material and methods: In a PLLA solution with or without (control group) fluvastatin, nanoparticles of HA or β -TCP were suspended, and CaP nanoparticle-dispersed PLLA sponge was fabricated. Fluvastatin was expected to be sustained-released from PLLA, as PLLA was degraded. In experimental groups, composite contained 1, 10, or 100 mg of fluvastatin. Under systemic anesthesia of 10-week-old female Wistar rats, abovementioned composites were applied to bone defect created at the tibiae (five for each group). Four weeks after the surgery, animals were sacrificed and then undecalcified ground sections were prepared. Finally, histological and histomorphometrical analyses were performed.

Results: No significant inflammatory response around the implants was indicated in all groups. In control group, abundant blood vessels and cell infiltration into interconnected pores of the materials were observed. In experimental groups, larger newly-formed bone was observed compared with that of control group. Newly formed bone was also seen to be in direct contact with the preexisting bone.

Conclusions and clinical implications: In this study, biodegradable polymer-CaP nanoparticle composite was observed to be biocompatible, without any inflammatory response. New bone formation was observed inside the composite, and fluvastatin seemed to stimulate the bone formation effectively. These results suggest that biodegradable polymer-CaP nanoparticle composite with fluvastatin is expected to be a potent bone substitute material.

339 Posters – Material Research

Functionality of a further developed implant system: mechanical integrity

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Background: Mechanical strength of the implant and implantabutment connection is crucial for predictable, long-term clinical results, and is always a target for continuous development.

Aim/Hypothesis: This study was set up to mechanically justify a further developed implant system compared to its forefather, a well-documented implant system that has been on the market for over 25 years.

Material and methods: The international standard ISO 14801: 2007 was employed. Implant-abutment assemblies of a further developed implant system (OsseoSpeed[™] EV, DENTSPLY Implants, Mölndal, Sweden) with both cylindrical and conical external shape, and with an internal conical abutment connection, were tested and compared with a control implant system (OsseoSpeed[™] TX, DENTSPLY Implants, Mölndal, Sweden) in similar configurations, lengths and diameters. Hence, seven different implant sizes and designs constituted the test group while five different sizes and designs constituted the control group. In order to limit the number of variables, the tests were conducted with similar abutment material and design (two component indexed titanium abutments from the same manufacturer). Test and control assemblies were tested at various loads during up to 5,000,000 cycles to comply with the ISO test standards. According to ISO 14,801, the fatigue limit was defined at the load level where three consecutive samples survived 5,000,000 cycles. Mechanical integrity analyses were done using Wöhler curves.

Results: The narrow 3.0 mm diameter implant assemblies resisted the lowest force, and moment, in both groups, although the narrow test implant assemblies were always superior (15%) in fatigue resistance compared to the narrow control assemblies. In fact, all test assemblies were between 11–20% superior in fatigue resistance (fatigue limit force and fatigue limit moment) compared to the corresponding control

assemblies in all dimensions. The widest diameter test implant-abutment assembly (5.4) resisted the highest force noted in the study.

Conclusions and clinical implications: It was concluded that all test assemblies with the further developed implant system outperformed the corresponding control assemblies. The implant system OsseoSpeedTM EV is mechanically justified for clinical use.

340 Posters – Material Research

Credibility of an up-dated implant system: implant-abutment leakage testing

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Background: Implant treatment is an area of continuous development of, for example, implant and abutment designs, fatigue strength, but also the widening of treatment indications. When further updating and improving the implant-abutment connection, the assurance of sealing of this interface is an important aspect for the clinical outcome.

Aim/Hypothesis: To study fluid leakage of the implant-abutment interface in an up-dated implant system with an internal conical connection.

Material and methods: To study the possible leakage through a conical seal interface per se, a narrow hole was drilled at the bottom of the implant. Secondly, a groove, parallel with the long axis of the abutment screw, was made on the threads of the abutment screw before mounting the implant-abutment specimen. Two-part abutments (TiDesign[™] EV, DENTSPLY Implants, Mölndal, Sweden) were tightened with 25 Ncm, to the implants (OsseoSpeed[™] EV 3.6S (M1.6) and 5.4S (M2.0), DENTSPLY Implants, Mölndal, Sweden). The fluid leakage test was performed under cyclic loading in accordance with the ISO 14801:2007 standardized method. Hence, the upper section of the specimens, were submerged in a container with physiological sodium chloride solution at room temperature simultaneously as the loading occurred. The lower section of the specimen engaging the apex of the implant (with the hole) was under diminished air pressure (-0.30 bars) and subsequently connected to a fluid level measurement pipe. Fluid leakage was evaluated by constant measuring of the fluid level in the measurement pipe giving a volumetric flow rate (Q = volume/time), during cyclic (1 Hz) loading of 100, 210 and 275 N, respectively. Possible leakage was evaluated from three specimens of either size, tightened with the set torque, after being loaded during 10 min. A reference implant system (OsseoSpeed[™] TX 3.5S/4.5, DENTSPLY Implants, Mölndal, Sweden) was also tested for fluid leakage.

Results: No leakage was detected for any of the test samples during the 10 min test, irrespective of degree of load, when the abutment was seated with the recommended torque. No implants from the reference groups posed any fluid leakage in the applied test environment.

Conclusions and clinical implications: The up-dated implantabutment connection can be considered a tight internal conical seal when the abutment screw is secured with 25 Ncm. The reference implant system, currently marketed, is equally tight and free from any fluid leakage in the internal junction between implant and abutment under the present test conditions.

341 Posters – Material Research

Effects of epigallocatechin-3-gallate on healing of the extraction socket with periapical lesion

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Background: The presence of inflammatory cells in extraction sockets with history of periapical lesions could be a risk factor for subsequent placement of dental implants or bone graft. Conventionally, the lesions were left to heal naturally for several months for resolution of any remaining sources of infection. During the healing period, dimensional changes in hard and soft tissues arise, and result in bucco-lingually inadequate bone volume for placement of dental implants and esthetically unpleasant prosthesis. Epigallocatechin-3-gallate (EGCG) is a major green tea catechin reported to have anti-inflammatory, anti-oxidative and anti-bacterial effects.

Aim/Hypothesis: This study aimed to evaluate the healing process of the extraction socket with periapical lesion following immediate graft with collagenated bovine bone mineral (CBBM) soaked with EGCG.

Material and methods: Mandibular second premolars of five male beagle dogs were used. In order to induce periapical lesions, dental pulp was necrotized and extirpated from each tooth then the root canals exposed to the oral cavity were covered with dental plaque. After 16 weeks of induction period, the teeth were extracted and bone grafting into the extraction sockets was performed. Groups were divided into three; control group with no treatment in the socket and test groups that received CBBM grafts in the socket with or without EGCG. 3D reconstruction and superimposition of the digital images were used to measure the changes in the alveolar ridge dimensions. Histologic and histometric analyses were performed. Statistical differences between measured parameters on buccal/lingual side in three groups were analysed with two-way ANOVA.

Results: In vertical dimension, buccal aspects reduced significantly compared to the lingual in all groups. Both test groups exhibited larger horizontal ridge width at the 4 mm level, compared to the control. Fibrosis and limited new bone formation were observed at the apical region of both test groups, where the extent of fibrosis was less in the CBBM + EGCG group than the CBBM group.

Conclusions and clinical implications: Within the limitations of this study, it can be concluded that bone regeneration at the

coronal region of the socket is not influenced by the presence of a periapical lesion; however, the healing at the apical region may occur by reparative fibrosis. EGCG could reduce the extent of fibrosis.

342 Posters – Material Research

Effects of Mg-ion and Ca-ion implantations on *Porphyromonas gingivalis* and *Fusobacterium nucleatum* attachment

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Background: Control of inflammation is important for the long-term implant success. Attachment of bacteria to the implant surface is the crucial step in the pathogenesis of periimplantitis, and must be investigated thoroughly to ensure the long-term stability and success of implants. Many researchers have reported inflammation problems related to implants of various surface types. However, the literature on correlations between ion-implanted surfaces and bacterial attachment, particularly as regards the effects of different ion types, is as yet insufficient.

Aim/Hypothesis: The purpose of this study was to evaluate the effects of ion implantation on *Porphyromonas gingivalis* (*P. gingivalis*) and *Fusobacterium nucleatum* (*F. nucleatum*) bacterial adhesion.

Material and methods: Titanium (Ti) discs of 15 mm diameter and 1 mm in thickness (n = 42, seven per group) were fabricated. Magnesium (Mg) and calcium (Ca) ions were implanted into the Ti surfaces using a plasma-source ion-implantation method. The surfaces' roughness, chemistry, morphology, and contact angle were analyzed by scanning electron microscopy, Rutherford back-scattering spectroscopy, Auger electron spectroscopy, and contact-angle meter. P. gingivalis and F. nucleatum strains were cultured under anaerobic conditions at 37°C for 72 h, in which 37°C suspension and all of the Ti specimens were immersed for 24 h. The specimens were examined under 1000X fluorescence microscopy magnification. The number and total area of bacteria in each of 10 separate fields were determined by computer imaging analysis. The data obtained were analyzed to assess the significance of the observed differences according to the method of surface treatment and ion implantation.

Results: The amounts of *P. gingivalis* and *F. nucleatum* attached to the Mg- and Ca-ion-implanted surfaces were greater than those attached to the non-implanted surfaces (P < 0.001). The types of ion and bacteria did not affect the amount of bacterial adhesion.

Conclusions and clinical implications: Attachments of *P. gingivalis* and *F. nucleatum* on ion-implanted surfaces were significantly larger and stronger than on control surfaces. The effect by ion type was subtle. Non-specific bonding derived from the electrostatic force effected by positively charged ions might be the predominant factor in adhesion. Additionally, the possibil-

ity of specific bonding was revealed in the case of Ca-ion implantation. Further investigations to clarify the bacterial adhesion mechanism are necessary.

343 Posters – Material Research

Hardness of new bone by bone grafting materials

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Background: Many kinds of bone grafts are used to replace or reconstruct skeletal critical sized defects resulting from periodontal disease, cyst, tumor and trauma. Autogenous bone has been most useful bone grafting materials because it is nonrejection, osteoinduction, bone conduction and osteogenic.

Aim/Hypothesis: The graft by autogenous bone has several disadvantages such as limitation of collection volume, operation is complicated and infection associated with harvesting bone. Many clinical reseaches of various bone grafting materials have reported. And, calcium phosphate have researched by reason of these are similar to mineral of mammals tooth and bone. In this study, the usefulness of bone grafting materials was evaluated *in vivo*.

Material and methods: Bone grafting materials were prepared with 4 different types; calcium phosphate (HAP, two types of β -TCP) and xenogeneic bone. Evaluation of their physical property were performed by XRD, FT-IR and SEM. Male Wister Rats were used for investigate. All rats were divided into six groups; HAP treated group, two types of β -TCP treated group, xenogeneic bone treated group, collagen treated group and control. One trephine detect, 9 mm diameter which was critical-size bone detect, was surgically created including the mid-sagittal suture. Then their bone formation modality were assessed by tissue section, bone formation volume by μ -CT and new bone hardness by nano indenter.

Results: Each materials was detected characteristic peaks by XRD and FT-IR respectively. The shape and the stomatal shapes were different by SEM. After 8 weeks, bone formation was confirmed in all groups other than no-filling group, and it was from the materials surface by u-CT and tissue section observation. The hardness of re-production bone were not inferior to pre-existing cranial bone.

Conclusions and clinical implications: It was suggested the final volume and quantity of new bone formation did not have the major difference in each bone filling material, and so artificial bone were useful for clinical case thinking in safe aspect.

344 Posters – Material Research

Effect of excimer ultraviolet to titanium modified by wire-type EDM and SLA

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Background: Titanium is major used for materials in dental implants because of high biocompatibility by the stable oxide layer. Some methods for surface treatment to get early osseointegration are reported and there are applied a clinic. The authors have demonstrated that wire-type electric discharge machining of titanium allowed a microstructured surface with an irregular morphology as well as thicker oxide layer. In addition, those surface characteristics have favorable scaffold properties for cell kinetics *in vivo* and bone formation by contactosteogenesis *in vivo*. Recently photocatalysis by radiation of ultraviolet lamp to improve titanium surface characteristic becomes the topic.

Aim/Hypothesis: This study aimed to investigate the surface characteristics at before and after radiation of ultraviolet lamps, excimer UV lamp (EX-UV) and the low-pressure Hg-UV lamp (Hg-UV), to titanium modified by wire-type electric discharge machining and SLA (sandblast and etching).

Material and methods: Two kinds of UV lamps, EX-UV and Hg-UV, were prepared and analyzed by measuring device of wavelength dispersion. Titanium plates $(10 \times 10 \times 1.0 \text{ mm})$ have surface modified by wire-type electric discharge machining (EDSurface) and machined surface were prepared at before and after radiation of UV. The surface of each specimen was characterized by measuring angle of contact of water, scanning electron microscopy (SEM), X-ray diffraction spectroscopy (XRD) and X-ray photoelectron spectroscopy (XPS). Each specimen was incubated in a-MEM containing 10%FBS with and without osteoblastic cells (MC3T3-E1). Adsorption of cell-binding proteins by each specimen was examined by XPS. Initial cell adhesion, proliferation and differentiation were measured by using WST-1.

Results: The peak of spectral distribution properties of EX-UV was observed at 175 nm. Whereas, the maim peak of Hg-UV was at 250 nm, and some small peaks were at other wavelength. An XPS and XRD survey indicated that the composition of the titanium oxide layer on EDSurface was consistent with machined surface. The adsorption of serum proteins on EDSurface was significantly higher than that with the another specimen. Both before and after radiation of each UV lamp, EDSurface showed super hydrophilia. The radiation of EX-UV lamp was able to increase contact angle as compared with Hg-UV on machined surface. In WST-1 assay, cell kinetics on EDSurface wrere better than machined surface.

Conclusions and clinical implications: It was found that there were major effects by radiation of EX-UV to machined surface. EDSurface showed high biocompatibility both before and after

radiation of UV. We will attempt analysis of surface chemical constitution change before and after the UV radiation, especially paying attention to oxide layer, and continue *in vivo* and *in vitro* study successively.

345 Posters – Material Research

In vitro precision of fit of screw-retained CAD/ CAM frameworks made from zirconium dioxide and titanium

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Background: Passive fit can minimize the risk of screw loosening in implant prostheses and prevent technical failures as well as biologic complications of the peri-implant tissues. Computer-aided design / computer-aided manufacturing (CAD/CAM) and laboratory scanners have introduced simplification of the digital workflow and new materials for long-span implant-bridges. However, the impact of the latest scanners and CAD/CAM systems on the precision of fit has not been sufficiently investigated.

Aim/Hypothesis: To analyze the precision of fit of screwretained long-span frameworks made from zirconium dioxide (ZrO) and titanium (TIT). The null-hypothesis was that the vertical microgap of ZrO frameworks using a laser and a tactile scanner would be the same between these two groups and there would be no difference compared to the titanium and the cast alloy frameworks.

Material and methods: A model case of an edentulous maxilla with six implants (FDI positions 15, 13, 11, 21, 23, 25) was fabricated from stable high quality acrylic material. The implants with a flat platform were placed parallel and with vertically aligned axes, respectively with 10° angulation for the anterior implants in the model. CAD/CAM ZrO frameworks (NobelProceraTM) for a screw-retained implant-supported 10-unit reconstructions were fabricated using a laser (ZrO-L, N = 6) and a mechanical scanner (ZrO-M, N = 5) for digitizing the implant platform and the cuspid-supporting framework resin pattern. Laser-scanned CAD/CAM titanium (TIT-L, N = 6) and cast CoCrW-alloy frameworks (Cast, N = 5) fabricated on the same model and designed similar to the ZrO frameworks were the control. The one-screw test (implant 25 screw-retained) was applied to assess the vertical microgap between implant and framework platform with a scanning electron microscope. The mean microgap was calculated from approximal and buccal values. Statistical comparison was performed with non-parametric tests.

Results: No statistically significant pairwise difference was observed between the relative effects of vertical microgap between ZrO-L (median 14 µm; 95% CI 10–26 µm), ZrO-M

(18 μ m; 12–27 μ m) and TIT-L (15 μ m; 6–18 μ m), while the values of Cast (236 μ m; 181–301 μ m) were significantly higher (*P* < 0.001) than the three CAD/CAM groups. A monotonous trend of increasing values from implant 23 to 15 was observed in all groups (ZrO-L, ZrO-M and Cast *P* < 0.001, TIT-L **Conclusions and clinical implications:** CAD/CAM technology using optical and tactile scanners allows for the fabrication of highly accurate long-span screw-retained implant-reconstructions made from zirconium dioxide and titanium. While the precision of fit of the cast alloy frameworks was clinically inacceptable, the titanium frameworks showed the most consistent precision.

346 Posters – Material Research

Micromobility of the implant/abutment interface for original and third-party abutments – a combined experimental and numerical study

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Background: The ongoing progress in CAD/CAM techniques in dentistry allows the construction of patient-individualised implant abutments. With the growing number of individualised CAD/CAM abutments, some of them specialised for different implant systems, the mechanical stability of the connection between implants and individualised abutments has to be studied.

Aim/Hypothesis: To determine the micro-mobility between implant and abutment for a small set of implants with prefabricated or individualised abutments in a combined experimental and numerical study.

Material and methods: Different CAD/CAM abutments on Straumann SLA Bone Level RC 4.1 implants were used: original Straumann abutments, third-party abutments from Astra Tech Atlantis and Nobel Biocare Procera. Mechanical tests were performed in two setups, both using optical methods to determine the relative implant/abutment movement. Resulting movements upon loading with 120 N and 500 N were registered with a resolution of 0.7 and 4.0 µm, respectively. Loading conditions were chosen following ISO 14801 (angulated loading direction of 30°, embedding implants 3 mm below nominal bone level, force application 8 mm above nominal bone level). For each implant/abutment combination, three specimens were measured. For a load of 120 N, measurements were repeated five times for every specimen. One of each implant/abutment combinations was scanned in a µCT scanner to create Finite-Element (FE) models. After validation using the experimental data, additional FE simulations were performed varying the loading direction and the bone height.

Results: At 120 N, the mean relative implant/abutment mobility was between 2 and 4 μ m for all abutment types (not significant for *P* = 0.05). Variance in the relative movement at 120 N for the third party abutments was more than twice as

high as for the original abutments. At 500 N, the original abutments showed a mean relative movement of 29 μ m, while the third-party abutments showed statistically significantly increased mobility (*P* = 0.05) of 36 and 51 μ m. The simulations showed movements similar to the experimental behaviour. Despite the conical design of the abutment neck, all models showed gaps to some extend between implant and abutment. Smallest gaps were found for the original abutments.

Conclusions and clinical implications: Depending on the magnitude of load applied, third-party abutments showed an increased variation in mobility (at 120 N) or a significantly increased mobility (at 500 N). Such an increased mobility can compromise the mechanical stability of implant supported prosthetic restorations, and may have a negative influence on the fatigue behaviour. Especially for single-tooth restorations the increased mobility should be taken into account. We would like to thank Straumann for providing materials.

347 Posters – Material Research

Experimental study of the wear behaviour of retentive attachment systems for removable partial dentures on endosseous dental implants

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Background: The long-term success of removable partial dentures as well as the patient's satisfaction with such dentures depend on a good mechanical connection between fixed and removable components and the ease of usage. Additionally, special care has to be taken to counter a possible non-parallel orientation of the implants that are used to anchor the dentures.

Aim/Hypothesis: It was the aim of this study to determine the wear behaviour and the change of retention force for two of two such attachment systems using different retention inserts with implant divergence of 0 and 10° in a mechanical simulation.

Material and methods: Two different attachment systems (Locator[®], Zest Anchor LLC, and SFI-Anchor[®], Cendres + Métaux SA) were used in this study, both with polymer retention inserts with forces of 3 N and 13 N. Additionally, a gold alloy retention insert was tested for the SFI-Anchor. Both attachment systems were mounted on the same implant type according to the manufacturer's instructions for use. For each combination of attachment system and retention insert, 20 specimens were placed in a custom wear simulation set-up, ten of them without and ten with a divergence of 10° between the orientation of the attachment system and the joining/separation direction of the experimental set-up. Each specimen was submitted to 5000 cycles of joining and separation the attachment system, using artifi-

cial saliva as lubricant. Forces during joining and separation were registered continuously, and the highest force during each separation phase was recorded. The influence of the wear on the surfaces was monitored using a scanning electron microscope.

Results: For the gold inserts, large variations could be observed within the same group. For the polymer-based retention inserts, within each attachment/insert group the specimens showed a consistent behaviour. The initial force levels were clearly above the nominal force level, especially for the groups with a divergence of 10° (up to 52 N for Locator with 13 N, 10°). The separation forces gradually decreased to the nominal force level within the first 300 (SFI-Anchor, 13 N, 10°) to 1500 (Locator, 13 N, 10°) separation cycles. The polymer SFI-Anchor insert showed almost no change in separation force after 1000 cycles. Overall, the 10° groups showed only a minor increased separation force compared to the 0° groups.

Conclusions and clinical implications: The retention forces of inserts for attachment systems are quiet predictable and controllable for the tested polymer-based retention elements. Especially for dentures on more than two implants the high initial force level has to be taken into account.

348 Posters – Material Research

The effect of mechanical and laser treatments on the titanium implant-adherent biofilms

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Background: The major cause of peri-implantitis is the attachment of biofilm on the surface of implant and it lead to marginal bone loss. For the treatment of peri-implantitis, it is essential to decontaminate the exposed implant fixture surfaces. Non-invasive treatments of peri-implantitis include manual debridement, ultrasonic debridement, air-abrasive device, laser treatment and photodynamic treatment

Aim/Hypothesis: This study aims to compare mechanical debridement with Er:YAG laser treatment in terms of capabilities to remove biofilm attached on the surfaces of dental implants *in vivo*.

Material and methods: Six Men were enrolled for this study. All volunteers were in sound medical and oral hygiene condition with physiological salivary flow rates of 1~1.5 ml/min and with no history of anti-biotic treatment for the past twelve months. Implant specimens (Megagen, Exfeel) were attached to palatal appliances, which were placed in the oral cavity of each volunteer for 3 days. Volunteers did not perform oral hygiene while they are wearing the appliances. After the study period, specimens were removed from appliances and broken down into four groups for biofilm removal between implant threads: control group, plastic curette debridement group, ultrasonic scaler debridement group and Er:YAG laser treatment group. After treatment, half of specimens underwent fixation and were evaluated using SEM. Live/dead cell assay has been performed for another half of specimens. The areas covered by dead cells (fluorescent red), viable cells (fluorescent green), and total cells were calculated as percentage of specific standard microscopic fields with the image analysis software.

Results: Er:YAG group showed unique surface morphology when evaluated with SEM. Live/dead cell assay showed no significant difference of live/dead cell ratio between plastic curette and ultrasonic scaler group, whereas Er:YAG laser group showed significant difference from other groups. (Live/dead cell ratio was significantly low for Er:YAG laser group.)

Conclusions and clinical implications: In terms of removal of biofilm between dental implant threads, this *in vivo* study showed that Er:YAG laser treatment is more efficient than mechanical debridement. The result of this study suggests that better accessibility of non-contact mode Er:YAG laser enabled more thorough debridement for dental implant threads than mechanical scaler does.

349 Posters – Material Research

In-office, immediate, and rapid preparation of autotooth biomaterial for alveolar bone reconstrunction

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Background: In order to preserve and restore tooth which plays a key role in mastication, pronunciation and esthetics, alveolar bone with good quality is an essential requirement. Therefore, there have been significant amount of studies and efforts to recover deficient alveolar bone as quickly, sufficiently and biologically as possible, and many graft materials for alveolar bone reconstruction have been introduced and are being already used clinically. Demineralized form of dentin can be more effective as a graft material. The ability of demineralized dentin to induce heterotopic bone formation is well established, but a conventional decalcification method takes time for 3-5 days and long treatment period may induce negative effects to various osteogenic proteins in dentin. Therefore, immediate bone grafting after extraction is impossible in this conventional method. In order for demineralized tooth of which excellence as graft material has been proven to be actively used clinically, therefore, a task that processing time has to be as shortened as to be used right after extraction has to be solved.

Aim/Hypothesis: Based on previous studies, we studied on the short processing time method with vacuum-ultrasonic machine in a clinic for single day surgery after extraction. The aim of this study was to evaluate the effect of using vacuum ultrasonic power on the demineralization and process of autogenous teeth biomaterial and study the clinical results of rapidly processed autotooth graft material in alveolar defects cases.

Material and methods: The present new method is that the extracted teeth with soft tissue trimming and pulp and hole formation were demineralized in 0.6 N HCl for 70 min by vacuum-ultrasonic accelerator with heat controlled. The characteristics of the processed teeth were evaluated by the scanning electron microscope features (SEM), energy dispersive x-ray spectroscopy (EDS) and western blot. For clinical trial, 103 patients of odontoma, dentigerous cyst, impacted third molar, apical periodontitis and implant cases were selected.

Results: The process could be finished within 2 h regardless of the form (powder or block). The EDS and the SEM were showed uniform demineralization in the autotooth biomaterial. Non-collagenous proteins were observed in western blot. We observed new bone formation in the histologic features and radiologic images. Clinical cases did not show any adverse response and the healing was favorable

Conclusions and clinical implications: The new processing method of autogenous teeth could make the immediate one-day graft possible after extraction.

350 Posters – Material Research

Histomorphometric analysis of implant-bone interface between SLA® and Joy 1 implant® in dogs

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Background: There are various methods for roughness of implant surface introduced to increase osseointegration of titanium implant. Newly developed Joy 1 implant®(DAS, Korea) enchanced implant osseointegration through SLA (sand blasted large-grit acid etched)

Aim/Hypothesis: The objective of this study was to compare the osseointegration of SLA® (ITI implant, Swiss) to these of Joy 1 implant®(DAS, Korea).

Material and methods: In 6 beagle dogs, 3rd and 4th mandibular premolar were extracted bilaterally. After 3 months, four transmucosal screw-shaped implants were placed in each mandibular edentulous premolar region. The implants have SLA (sandblasted, large-grit acid etched) implant surface. Control groups were SLA® (10 mm length, 4.1 mm width) and experimental groups were Joy 1 implant® (10 mm length, 4 mm width). The implants were allowed to heal for 4 or 8 weeks. At the end of the experiment, the dogs were sacrificed and each of implant sites was dissected and processed for histomorphometric analysis. The results of the bone-implant contact (BIC) and bone-ingrowth area (BI) were compared in SLA® and Joy 1 implant®. Statistical analyses were performed using Mann-Whitney U test and Wilcoxon test. **Results:** Histomorphometrical analysis showed direct osseous integration for both implants. Mean of BIC was $60.61 \pm 23.40\%$ at 4 weeks and $60.77 \pm 16.62\%$ at 8 weeks after implantation in SLA® implant, whereas mean values of Joy 1 implant® was $50.26 \pm 28.47\%$ and $59.23 \pm 4.17\%$ at corresponding time intervals. Mean of BI was $60.61 \pm 23.40\%$ at 4 weeks and $60.77 \pm 16.62\%$ at 8 weeks after implantation in SLA® implant, whereas mean values of Joy 1 implant® was $50.26 \pm 28.47\%$ and $59.23 \pm 4.17\%$ at corresponding time intervals. Mean of BI was $60.61 \pm 23.40\%$ at 4 weeks and $60.77 \pm 16.62\%$ at 8 weeks after implantation in SLA® implant, whereas mean values of Joy 1 implant® was $50.26 \pm 28.47\%$ and $59.23 \pm 4.17\%$ at corresponding time intervals. No significant difference between both types of implants could be detected after 4 and 8 weeks healing periods in BIC and BI.

Conclusions and clinical implications: The results indicate that there was no difference in osseointegration between SLA® implant and Joy 1 implant® regarding BIC and BI, Joy 1 implant® showed similar histological response with SLA® implant.

351 Posters – Material Research

Comparison of biofilm on titanium and zirconia surfaces: *in vivo* study

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Background: As osseointegrated implants are more widely used, studies about implant abutment materials are also on progress. As biofilm is the main factor of peri-implantitis, the main reason of implant failure, studies about biofilm formation on implant abutment will be highly meaningful. Recently, zirconia as well as titanium, is drawing attention as a new implant abutment due to its aesthetic and anti-microbial action.

Aim/Hypothesis: The aim of the present study was to compare the biofilm formation on titanium and zirconia surfaces *in vivo*.

Material and methods: For the biofilm formation on titanium and zirconia in oral cavity, after producing oral appliances using acrylic resin and orthodontic wire tailored to 9 subjects. Titanium and zirconia specimens was made (6 mm \times 6 mm \times 2 mm), and fixed them on oral appliances and maintained them in oral cavity of testy subjects for 24 and 72 h. Test subjects who have equipped two pairs of specimens maintained oral hygiene not by using toothpaste but only tooth brushing. After 24 and 72 h, we removed and observed specimens through SEM.

Results: Biofilm formation showed large deviation depending on individuals. For formation comparison between titanium and zirconia for 24 h, zirconia showed less biofilm formation than titanium. Biofilm formation showed large deviation depending on individuals. As for formation comparison between zirconia and titanium, the degree of biofilm formation in zirconia was less than it was in titanium after a lapse of 24 h. The results of biofilm formation in 72 h trial show that zirconia has a inclination to formate less biofilm than it was in titanium. **Conclusions and clinical implications:** Based on above results, we can conclude that early biofilm attachement happens more easier in titanium than in zirconia.

352 Posters – Material Research

Laboratory analysis of the removal torque and the interface implant/abutment in taper morse implants after mechanical cycling

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Background: The success of the treatment with dental implants depends on many factors affecting the bone-implant, implant-abutment and abutment-prosthesis interfaces. The restorations on the implant are continually subject to the forces of separation of the screw joint, and these include excursive contacts, eccentric contacts, interproximal contacts, contacts and structures in cantilevers not passive. Thus, to minimize the forces that tend to separate the parts joined by screws, loosening or fracturing them, it was recommended to avoid excessive angulation of the implant restorations with cantilever, contacts centric and non-axial structures which do not present a passive fit.

Aim/Hypothesis: The objective was to evaluate the mechanical behavior and the interface abutment/implant in implants with Morse taper connection after application of cyclic loads.

Material and methods: A total of 30 implants Morse taper (Implacil De Bortoli, Sáo Paulo, Brazil) diameter of 4 mm and length of 11 mm and 30 straight solid pillars, divided into two groups: control group (Gcon) (n = 15), where the sets (abutment / implant) received only the torque of 25 N, and were evaluated; experimental group (Gexp) (n = 15), where the assemblies were torqued and subjected to cycles of mechanical fatigue at a load of 100 N, frequency of 4 Hz and 360,000 cycles. In each group of ten implants the abutments were removed and the removal torque values were recorded. Other five in each group were embedded in resin metallographic after the respective tests and cut along the perpendicular center axis of the contact sets for analysis of the internal walls between abutment and implant.

Results: The Gexp group had an average increase in the removal torque value of 19.7% (29.9 N) while the group Gcon an average decrease in removal torque value of 11.8% (22.3 N), indicating a statistically significant difference ($\alpha = 0.05$). In the interface of the assemblies Gcon group presented different values in the positions evaluated, different from group Gexp where space after cycling was no longer visible even in large increments.

Conclusions and clinical implications: After application of cyclic loading on the implants, they showed an increase in the amount of removal torque. It was also observed through the metallographic sections, an increase in contact between the inner walls of the sets after application of loads.

Development and '*in vitro*' biological assessment of gelatin-silica coatings

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Background: In spite of the great results obtained in the osseointegration of Ti implants, there are still some aspects that should be enhanced, for instance, time for total stability and integration in the jaw of patients with health troubles. Silica based biomaterials are currently investigated by our research group as coatings for dental implants due to their biocompatibility and because they release Si ions which promote osteoblast differentiation and enhance osteocompatibility. Previous *'in vivo'* results showed a great improvement and a faster osseointegration than for titanium implants without coating.

Aim/Hypothesis: With the aim of continuing to improve the properties of these materials, gelatin will be added because of its cell adhesion ability and consequently its osteoregenerative properties. Therefore, the aim of this work is to develop a silica-gelatin hybrid coating for dental implants with tailorable degradation and properties, to improve the osseointegration of the traditional titanium implants.

Material and methods: Coatings were synthesized by the solgel method from specific mixtures of different alcoxysilanes and gelatin. The degradation of film was determined by the weight loss as a function of immersion time in distilled water at 37°C. The release of silicon and gelatin were also monitored over time. To analyze '*in vitro*' behavior, Alizarin Red Test was carried out to determine the presence of deposited calcium. For '*in vivo*' tests, Ti screws coated with the sol-gel film were implanted in rabbit tibias. The histology around the explanted samples will be studied and evaluated by different means.

Results: Degradation kinetic showed that coating with the highest content in gelatin degrade more rapidly, which is in good agreement with the results of the silicon release assay. The *'in vitro'* assay showed that in 7 days, the mineralization values of our coatings are not significantly different from the Titanium Control and Plastic Control. However, for the period of 21 days, the enhancing effect of the gelatin was noticeable, being the material with the highest content the one with the highest values of mineralization.

Conclusions and clinical implications: The results of this study are encouraging for the use of these silica-gelatin hybrids as bioabsorbable and osseoinductive coatings for dental implants. *'in vivo'* studies are being carried out and results could be shown in the conference. In future work the ability of these coatings to form hydroxyapatite in SBF, as well as the capacity

to be used as controlled release systems of therapeutic agents, will be evaluated.

354 Posters – Material Research

Report on characteristics of abutments made with cobalt-chromium alloy

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Background: Cobalt-chromium alloy has a long history as a dental prosthesis material. However, most literature and reports assumed that we use cobalt-chromium alloy as a superstructure. Few reports assumed that we used it as an abutment. Today besides bridges (ex. FPB) we also use it in combination with an implant body (fixture). When materials of the implant body are different from an abutment, the issue of corrosion by galvanic electric potential is about a concern. Therefore studied an abutment fabricated in cobalt-chromium alloy and looked into its when used in combination with an implant body made of different material.

Aim/Hypothesis: The aim of this study is to evaluate release of metal ions and chemical character when we used a cobalt-chromium alloy abutment in combination with an implant body made of a different material (CP-Ti).

Material and methods: Six abutments were designed and fabricated by CAD/CAM using a wrought cobalt-chromium alloy disk (CCA-002D, GC). Three abutments were attached to implant bodies (GC Implant Aadva ϕ 4.0 × 10 mm, CP-Ti Grade 4, GC) respectively. Abutments were retained with abutment screws (Ti-6Al-4V, GC) tightened to 20 N/cm with an electronic torque controller. Samples were immersed in 0.9% NaCl and 1% lactic acid solution (pH = 2.3, 37 ± 1°C). Released metal ions were measured using ICP-AES (ICPS-7000, SHIMADZU). Tests were conducted according to ISO10271:2001(Dental metallic materials – Corrosion test methods). Amount of released metal ions were compared to each condition and subjected to statistical analysis (Student's *t*-test).

Results: In evaluation of the release of metal ions in the 0.9% NaCl and 1% lactic acid solution, the quantity of the metal ions which are released from abutment in combination with implant bodies (CCA-002D with GC Implant Aadva) was lower than that of abutments immersed separately (CCA-002D) (P < 0.01).

Conclusions and clinical implications: When an abutment made of cobalt-chromium alloy is attached to an implant body made of pure titanium, the release of metal ions from the abutment decreased. This could be caused by polarization created by different metal contact. As a result of these tests, abutments made of cobalt-chromium alloy milled by CAD/CAM may have potential in clinical situations involving implant dentistry, and expected to show good prognoses. Biological tests are currently being conducted and will also be reported.

Comparison of color gradation of natural central incisors and corresponding CAD/CAM restorative materials

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Background: CAD/CAM restorations have become a standard procedure in prosthodontic rehabilitation. The color of natural teeth is complex, not monotone, and knowledge of tooth color distribution is essential for esthetic implant restorations.

Aim/Hypothesis: This study aimed to analyze the color gradation of natural maxillary central incisors and full-contour CAD/CAM restorative materials by measuring three regions from incisal to gingival.

Material and methods: The following three full-contour CAD/ CAM restorative materials were tested: IPS Empress® CAD LT (Ivoclar Vivadent), 16 shades; IPS e.max® CAD LT (Ivoclar Vivadent), 13 shades; and IPS Empress® Multi (Ivoclar Vivadent), five shades. The color of 34 all-ceramic crowns milled from CAD/CAM restorative materials and natural maxillary central incisors of 300 individuals was measured using a spectrophotometer (Crystaleye Spectrophotometer®; Olympus) and the CIE color coordinates L*, a*, and b* in three regions of each tooth/crown (cervical, body, and incisal) were determined. Color differences (dE) between the all-ceramic crowns and natural teeth were calculated. Analysis of variance and Scheffe's multiple comparison tests were used for statistical analysis.

Results: Sixty-seven percent of the 300 natural teeth matched the all-ceramic crowns (dE < 3.0). dE values between full-contour CAD/CAM all-ceramic crowns and natural central incisors were 2.1 ± 0.1 , 1.8 ± 0.1 , and 2.1 ± 0.2 in the cervical, body, and incisal areas, respectively. No significant differences in mean dE values were observed among the three areas.

Conclusions and clinical implications: Natural teeth exhibited a good color match (dE < 3.0) with the corresponding fullcontour CAD/CAM restorative materials. Analysis of color gradation data from the incisal to the cervical regions of a tooth is useful for selecting appropriate CAD/CAM restorative materials.

356 Posters – Material Research

Nanoporous surface of titanium enhances implant osseointegration

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Background: Titanium implants surfaces are commonly gritblasted and acid-etched. However, the obtained micro rough surfaces are not controlled at the nanometer range for specific interaction with proteins and cell focal adhesion complexes. It has been recently shown that nanostructures on titanium implants significantly enhanced adhesion and osteogenic differentiation of mesenchymal stem cells (Lavenus S. et al. Eur Cells & Materials, 2011). In good agreement with *in vitro* results, bone apposition and bone strength were enhanced by nanostructured implants in tibia of rats (Lavenus S. et al. Nanomedicine, 2012).

Aim/Hypothesis: This study aims at comparing the osseointegration of machined, standard large grit-blasted acid-etched (GBAE) and nanostructured (NANO) implants in femur of rabbits.

Material and methods: Three groups of titanium cylindrical implants (4.2 × 6 mm) having an apical bone growth chamber were prepared. GBAE surface was blasted with large alumina particles and etched in a mixture of sulfuric and hydrochloric acids. Nanostructures were made by anodization in a hydrofluoric solution. Surface morphology was analyzed by FEG-SEM. After receiving ethical approval, 10 female adult NZW rabbits (Charles River Lab) were operated under general anesthesia. Implants were press-fitted in both femoral epiphyses. Pull-out testing and histomorphometric analysis were performed on explants. The bone implant contact (% BIC) and bone growth in the chambers (% BG) were measured by image analysis (ImageJ). Statistic tests were performed by ANOVA.

Results: SEM images show contiguous titania nanotubes of 40 nm in diameter covering entirely the NANO surface. Typical macro and micro rough surface was observed in the GBAE group. After healing for 4 weeks, pull-out testing gave higher values for the NANO than for other groups. Histomorphometric analysis showed that bone was in direct contact with the NANO surface while fibrous tissue was still visible around other types of implants. Both the BIC and BG values were higher for the NANO than for other surfaces.

Conclusions and clinical implications: This study shows that the nanostructured titanium surface improved the osseointegration of titanium implants. These preliminary results should be confirmed with commercial dental implants in pre- and clinical situations.

357 Posters – Material Research

Primary stability of different implants placed in standardized porcine bone cylinders: insertion torque and resonance frequency analyses

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Background: Primary stability is achieved when the implant is positioned into the host bone site and there is a direct mechanical contact between its surface and the surrounding bone. The achievement and maintenance of primary implant

stability is one important prerequisite for successful clinical outcome of immediate loaded implants.

Aim/Hypothesis: This study evaluates the primary stability of two different implants, placed in standardized porcine bone cylinders of high and low density, analyzing Insertion Torque (IT) and Resonance Frequency Analysis (RFA).

Material and methods: Porcine bone cylinders were removed from the femur head (high density bone - HDB) or from the mandibular condyle (low density bone - LDB). The cylinders standardization was certified by digital 2D radiographic analysis of bone density: cylinders with radiographic 2D bone densities values \geq 105 were selected as HDB, and with values \leq 95 were selected as LDB. Cylinders with radiographic density values between 96 and 104 were discharged. A total of 20 bone cylinders were certified (10 of HDB and 10 of LDB). Then, twenty Neodent® implants of two different models (both of them, according to the manufacturer, are indicated for insertion in bone types III and IV) were divided in four groups: Group 1 (G1) - 5 Drive CM implants placed in five HDB cylinders; Group 2 (G2) - 5 Drive CM implants placed in 5 LDB cylinders; Group 3 (G3) - 5 Alvim CM implants placed in 5 HDB cylinders; Group 4 (G4) - 5 Alvim CM implants placed in five LDB cylinders. During each implant placement the insertion torque was recorded. Following implant installation, RFA was measured with the Osstell® in four different directions (with 90° of difference among them); the average of the four implant stability quotient (ISQ) values was considered the implant ISQ value.

Results: The mean \pm standard deviation of IT values (N.cm) were: G1 = 47.66 \pm 8.51; G2 = 18.46 \pm 4.15; G3 = 38.18 \pm 2.75; G4 = 21.00 \pm 9.43, with statistically significant differences between G1 vs. G2 and G1 vs. G4. The mean \pm standard deviation of ISQ values were: G1 = 58.40 \pm 17.64; G2 = 32.90 \pm 4.51; G3 = 55.45 \pm 17.43; G4 = 27.40 \pm 8.75, with statistically significant differences between G1 vs. G4 and G3 vs. G4 (for all analyses, Kruskal Wallis + Dunn tests, P < 0.05).

Conclusions and clinical implications: Drive CM implants showed better numerically results in biomechanical assays, but the differences between the two types of implants for installation in the same type of bone (LDB or HDB) cylinders were not statistically significant.

358 Posters – Material Research

Osteoblast cell adhesion and spreading on nitrided titanium surface

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Background: Osteoblast attachment on titanium implant surfaces is essential to establish a successful osseointegration. Thus, several surface functionalization strategies have been developed to stimulate a fast cellular response to enhance the initial bone formation. **Aim/Hypothesis:** The aim of this study was to evaluate the initial cell response to a new titanium surface modified by cold plasma nitriding.

Material and methods: Moderately rough (Sa $1.0 \pm 0.1 \ \mu m$) grade 4 titanium discs $(12.7 \times 2 \text{ mm})$ were cleaned with argon to remove contaminants and nitrogen ions were bombarded onto the surface using cold plasma (TiN). The control group received no additional treatment (Ti). The topography was evaluated by atomic force microscopy, the chemical profile was determined by x-ray photoelectron spectroscopy and surface wettability was calculated by water contact angle measurement. Human osteoblasts (SAOS-2) were seeded on sterile discs (n = 6) and cultured for 24 h to evaluate the effect of the surface treatment on cellular response. The initial cell adhesion and proliferation were evaluated by tetrazolium colorimetric assay and cell spreading was investigated by scanning electron microscopy (SEM). Confocal laser scanning microscopy was used to evaluate cell morphology after staining with DRAC5 and phalloidin dyes.

Results: TiN group exhibited more nanostructures, higher nitrogen content in the surface and lower water contact angles $(75.6 \pm 3.1^{\circ})$ compared to Ti group $(80.4 \pm 4.6^{\circ})$. After 24 h, TiN surface exhibited more cells attached and early proliferation rate than Ti group (P < 0.05). SEM images revealed a higher level of cell spreading occurring on TiN surface. Moreover, wider osteoblasts with more extended protrusions were seen on TiN surface at the confocal images.

Conclusions and clinical implications: The present results suggest that nitriding the surface with cold plasma can be a useful tool to improve cell adhesion on titanium implants.

359 Posters – Material Research

Influence of Ti-coating on the osseointegrative properties of polyetheretherketone (PEEK) implants: Preliminary results of a pilot study in sheep

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Background: Long-term success of endosseous implants has continuously improved over the last decades, such that osseointegration has been considerably refined by appropriate implant designs and implant surface improvement. Beneath titanium and zirconia recently also polyetheretherketone (PEEK) has been promoted as suitable implant material especially in orthopedics. Yet, since PEEK offers limited possibilities of direct bone anchorage, special coatings like e.g. titanium could help to improve osseointegration.

Aim/Hypothesis: The aim of the present in-vivo study was to analyze the influence of a Ti-coating on the osseointegrative

properties of standard and reinforced polyetheretherketone (PEEK) implants compared to uncoated PEEK implants surfaces. The H1-hypothesis was that Ti-coated PEEK implants were expected to enhance bone ongrowth and thus device fixation-retention.

Material and methods: Four materials were tested: uncoated PEEK (PEEK), uncoated carbon-fiber reinforced PEEK (CFR-PEEK), low-roughness titanium-coated PEEK, high-roughness titanium-coated CFR-PEEK. Coatings were applied by means of plasma spraying. *in vivo* model: In n = 6 sheep implants were bilaterally placed in iliac shafts (n = 3 per surface group per animal). After 2 and 12 weeks n = 3 implants per group were examined histologically (bone-to-implant-contact) and n = 6 implants per group underwent by pull-out test. Additionally, a histomorphometrical and fluorescent microscopic analysis were performed.

Results: All implants could be rapidly placed, without complications and with good primary stability. Biomechanical pullout-testing after 12 weeks revealed statistically significant $(P \le 0.001)$ increased retention of low-roughness titaniumcoated PEEK (820 \pm 200 N) and high-roughness titaniumcoated CFR-PEEK (1180 \pm 330 N) compared to uncoated PEEK $(30 \pm 19 \text{ N})$ and uncoated CFR-PEEK $(39 \pm 24 \text{ N})$. Additionally, a statistically significant increase $(P \le 0.001)$ of values from 2 to 12 weeks of low-roughness titanium-coated PEEK (2 weeks: 228 ± 77 N) and high-roughness titanium-coated CFR-PEEK (2 weeks: 330 ± 110 N) could be detected. Both uncoated PEEK materials exhibited no striking differences between 2 and 12 weeks. Qualitative microscopic evaluation demonstrated new bone formation adjacent to the implant surface in each group. After 12 weeks both coated implants groups showed an increase of the bone-to-implant-contact. Histomorphometrically, a percentage increase of new bone formation with time was observed. Fluorescent labeling confirmed the histological and biomechanical findings.

Conclusions and clinical implications: In comparison to uncoated PEEK, surface coated with plasma sprayed titanium revealed highly favorable biomechanical and biological characteristics for standard as well as carbon-fiber reinforced PEEK. For a possible application in implant dentistry an additional analysis in a suitable soft tissue model is necessary.

360 Posters – Material Research

Effect of abutment screw design on the seal performance of an external hex implant system

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Background: Seal integrity of the implant-abutment-junction (IAJ) has significant clinical relevance due to the potential detriments associated with an inferior seal such as microleakage. Abutment screw design is a paramount factor as the screw generates the preload required to establish and maintain system stability.

Aim/Hypothesis: To characterize the IAJ seal robustness of an external hex implant system subjected to dynamic loading with titanium and Gold-Tite[®] abutment screws.

Material and methods: The apex of the 4 mm test implants was modified to have a barb fitting and through hole. The implants were embedded in a phenolic block 3 mm supragingival, and the implant barb was connected to a 7 psi peristaltic pump containing red dye. A GingiHue[®] abutment and test screw were assembled with 20 N-cm of torque. The block was mounted at 20° off-axis in a clear water tank, and the IAJ was magnified 50x with a video camera. The system was cyclically loaded at 100 N for 100 k cycles at 30 Hz. Following this wear phase, the pump was activated and the frequency was reduced to 2 Hz for 1k cycles to monitor the IAJ. The wear-monitor routines were incremented in 50 N load steps until a breach occurred, and the system components were then examined to detect yield and/or fracture damage. Implant systems with titanium screws (n = 5) and Gold-Tite screws (n = 5) were assessed.

Results: Statistical significance (P < 0.05) was detected between the two groups. The mean seal strength of the implant systems tested with the Gold-Tite screw was approximately 35% greater than that of the implant systems utilizing the titanium screw.

Conclusions and clinical implications: The Gold-Tite abutment screw provides a significant improvement in the seal robustness of an external hex implant system subjected to clinically relevant loading conditions.

361 Posters – Material Research

Initial adhesion on nanoscale features of the titanium surface: effects of deposition time in NaOH solution

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Background: The present study aimed to investigate cellular behavior on nanoscale features of a titanium surface by controlling the deposition time in NaOH.

Aim/Hypothesis: We then wanted to evaluate the influence of titanium at nanosheet surface on initial attachment of rat bone marrow cells to potentially increase the success rate of titanium implants.

Material and methods: Titanium disks were untreated or soaked in 10 mol/l NaOH for 5 min, and 1, 3, 9 and 24 h. SEM and SPM were used to evaluate the nanoscale features. Rat bone marrow cells were seeded on the samples in osteogenic differentiation medium. It was measured adhesion behavior of cow's serum albumin and fibronectin by using BCA protein assay Kit. SD rat bone marrow cells (4×104 cells/well) were plated into each well, and the number of cells were counted at 1, 3, 6, 24 h of culture by using CellTiter-BlueTM Viabillity Assay Kit.

Results: Nanofeatures were detected at 1 h after NaOH treatment and were well established at 9 h. The adhesion number of both proteins on Ti surface soaked in 9 h and 24 h was significantly higher than that of other groups. After 1 and 3 h of culture, the number of cells on Ti surface soaked in 9 h and 24 h shows significantly more than those of the other groups, but after 6 and 24 h of culture, no significant difference was observed between all groups

Conclusions and clinical implications: We found that the nanoscale modification of a titanium implant surface by NaOH treatment affects initial adhesion of rat bone marrow cells and enhances mineralization. Further development of advanced implant materials using nanotechnology may improve osseointegration.

362 Posters – Material Research

Evaluation of three different validation procedures regarding the accuracy of templateguided implant placement: an *in vitro* study

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Background: Appropriate validation procedures have been developed and used for evaluating the implementation accuracy of navigated implant systems. The validation procedures serve for the illustration and the quantification of deviations from the preoperatively planned to the postoperatively achieved implant positions. Implementation errors in clinical and experimental use may be associated with various causes such as scanning, processing, surgery and prosthetics. As yet, none of the studies known to the authors has assessed the influence of validation procedures on the calculation of implementation accuracy.

Aim/Hypothesis: For this reason this experimental study was firstly aimed to evaluate the implementation accuracy of the NobelGuideTM concept avoiding clinical impact parameters and secondly to validate two validation procedures vs. a reference method.

Material and methods: Overall, 60 implants were placed in 10 artificial edentulous mandibles according to the NobelGuideTM protocol. For merging the pre- and postoperative DICOM data sets three different fusion methods (Triple Scan Technique, NobelGuideTM Validation software and AMIRA[®] software as reference) were applied. Discrepancies between the virtual and the actual implant positions were measured.

Results: The mean deviations measured with AMIRA[®] were 0.49 mm (implant shoulder), 0.69 mm (implant apex) and 1.98°mm (implant axis). The Triple Scan Technique as well as the NobelGuideTM Validation software revealed similar deviations compared to the reference method. A significant correlation between angular and apical deviations was seen (r = 0.53; P < 0.001). A greater implant diameter was associated with greater deviations (P = 0.03).

Conclusions and clinical implications: The Triple Scan Technique as a system-independent validation procedure as well as the NobelGuideTM Validation software are in accordance with the AMIRA[®] software. The NobelGuideTM system showed similar or less spatial and angular deviations compared to others.

363 Posters – Material Research

Comparison of IL-1 β & TNF- α in peri-implant crevicular fluid around zirconium healing abutments with or without peri-implant mucositis

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Background: Zirconium dioxide are currently widely used as permucosal implant abutments because of excellent aethstics. However, the responses of zirconium dioxide to peri-implant soft tissues are scarce.

Aim/Hypothesis: The purpose of this study was to investigate IL-1 β and TNF- α in peri-implant crevicular fluid around zirconium and titanium healing abutments with or without periimplant mucositis in dogs.

Material and methods: The third and fourth mandibular premolars and the first mandibular molar of three beagle dogs were extracted bilaterally. Eight implants were inserted each dog after healing. Two months later, the zirconium and titanium healing abutments were connected. Plauqe index, gingival index, pocket probing depth and peri-implant crevicular fluid were collected as the initial time. Then ligaments were placed around the neck of healing abutments on one side to induce peri-implantitis. Four weeks later, plauqe index, gingival index, pocket probing depth and peri-implant crevicular fluid were collected again. Plauqe index, gingival index, pocket probing depth, the volume of peri-implant crevicular fluid and IL-1 β and TNF- α in peri-implant crevicular fluid of zirconium and titanium healing abutments with or without peri-implant mucositis were analyzed.

Results: Plauqe index, gingival index, pocket probing depth, the volume of peri-implant crevicular fluid, the level of IL-1 β and TNF- α in peri-implant crevicular fluid around zirconia and titanium healing abutments after ligation in inflammation side and uninflammation side were similar. But all the indexes are higher in inflammation side than uninflammation side, and there were statistical differences between inflammation side and uninflammation side.

Conclusions and clinical implications: Zirconium dioxide had a similar response to peri-implant soft tissue compared to Titanium. Zirconium dioxide may be promising as dental implant abutment materials.

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